Bioengineering Research Grants (BRG) New Awards Fiscal Year 2004

Grant:	1R01CA109754-01	
Principal Investigator:	ACHILEFU, SAMUEL	PHD
Title:	Optical Probes & Methods for Imaging Integrin Expression	
Institution:	WASHINGTON UNIVERSITY	ST LOUIS, MO
Project Period:	2004/09/21-2009/08/31	

DESCRIPTION (provided by applicant): Integrins are adhesion molecules associated with several hallmarks of malignant tumors including tumor invasion, formation of metastasis, tumor-induced angiogenesis and apoptosis of aberrant and activated endothelial cells. Expression in tumor cells of ava3integrin (ABIR) alters the interaction of cells with the extra-cellular matrix, and accelerates tumor growth and invasiveness. Due to involvement with these critical stages of tumor progression, integrins are attractive target for molecular level strategies aimed at imaging and treating pathogenesis. A common recognition motif for integrins is a structurally diverse tripeptide sequence, RGD (Arg-Gly-Asp). Analogues of RGD peptides (RGDPAs) provide an approach to target integrin expression. Highly sensitive, non-ionizing diffuse optical tomography methods can be used to image the distribution of target integrin in diseased tissue. Our preliminary data show that conjugating RGD-containing peptides to near infrared (NIR) fluorophores produces compounds that specifically target ABIR-positive tumors. Accordingly, we plan to (1) use computational methods to design, screen, select, and optimize 60 ABIR-avid NIR-RGDPAs; (2) develop method for synthesizing the peptides and their conjugates; (3) evaluate the ABIR binding affinity, internalization, cytotoxicity, proliferative effects, and subcellular distribution of NIR-RGDPAs in cells by in vitro assays; (4) develop and use a diffuse optical tomography system to evaluate the localization of candidate probes in vivo; and (5) evaluate the biodistribution of selected cypate-RGDPAs and optimize dosage for in vivo use with ex vivo fluorescence. The immediate goal for the first year is to assemble a highly sensitive DOT system, select bioactive NIR-RGDPAs by computational screening and initiate their synthesis. The long term goal is to develop a technology platform for rapid screening of bioactive molecules and for contrast-mediated molecular optical imaging of ABIR-positive diseases and neovasculature. At the completion of this project, we will (1) identify at least one ABIR-specific optical probe for translational research, (2) develop software for "virtual screening" of putative bioactive ligands, and (3) develop a DOT system for longitudinal imaging of drug distribution in animals. All these products will be made available to other investigators.

Grant:	1R01DK063123-01A2	
Principal Investigator:	AMEER, GUILLERMO A	BS
Title:	A system to remove b2-microglobulin from blood	
Institution:	NORTHWESTERN UNIVERSITY	EVANSTON, IL
Project Period:	2004/07/01-2008/05/31	

DESCRIPTION (provided by applicant): The overall goal of this proposal is to develop a blood detoxification system that is specific for b2-microglobulin (b2m), an amyloidgenic protein that has been implicated in dialysis-related amyloidosis (DRA). DRA is an incapacitating, potentially fatal, and unavoidable consequence of long-term renal failure and the inability of current medical technology to replace all aspects of kidney function. The lack of b2m-clearance by the kidneys results in elevated plasma concentrations of this protein, followed by b2m-amyloid deposition in tissues via mechanisms that are not fully understood. Although several attempts have been made to reduce b2m plasma concentrations, the incidence and complications of this disease remain a significant problem. It is proposed to investigate an extracorporeal, immunoadsorptive approach that uses immobilized recombinant single-chain antibody fragments to remove b2m. Single-chain variable region antibody fragments (scFv) against human b2m will be produced using molecular biology and fermentation techniques, characterized and immobilized onto a porous support for use in a novel adsorber design. The efficacy of the adsorber will be tested in blood in vitro and its biocompatibility will be tested ex vivo in sheep. The results of the proposed research could potentially lead to the following: 1) use of a recombinant antibody fragment immunoadsorber as a therapy to slow the progression of DRA while avoiding the indiscriminate loss of needed proteins and 2) use of specifically designed immunoadsorbers as tool to help elucidate the role of suspected middle molecular weight soluble factors in the morbidity of end stage renal disease. Achieving proof of principle using DRA as a model disease would support the tailored application of this technology for the removal of target molecules that cause other pathologic states.

Grant:	2R01AR043628-10	
Principal Investigator:	ATESHIAN, GERARD A.	PHD
Title:	Biotribology of Diarthrodial Joints	
Institution:	COLUMBIA UNIV NEW YORK MORNINGSIDE	NEW YORK, NY
Project Period:	1995/06/15-2008/04/30	

DESCRIPTION (provided by applicant): The primary function of articular cartilage is to serve as a bearing material for diarthrodial joints. As part of our recent studies, we have demonstrated from theory and experiments that cartilage can provide efficient lubrication even under boundary friction, primarily because its interstitial water may pressurize considerably under loading. In this application, we propose to follow-up on these findings by addressing specific questions related to the functional effectiveness of the lubrication mechanism in diarthrodial joints: a) Since the friction coefficient is observed to increase over time to functionally detrimental values under laboratory testing conditions, what makes it stay sufficiently low under physiological loading conditions in situ? b) How effective are the boundary lubricants in synovial fluid, after accounting for the role of interstitial fluid load support? c) Is there a functionally significant mixed lubrication regime in the initial response to joint loading during which lubricant is temporarily trapped between articular surfaces, and if so, does the duration of this mixed regime depend on the type of lubricant? While it may be attractive from the clinical and commercial perspective to consider joint lubrication in terms of a phenomenon arising from synovial fluid and its constituents (e.g., intra-articular injection of hyaluronan and other "lubricants"), their relative contribution to the low frictional properties of articular cartilage are not well understood. With an original theoretical framework, validated using novel experimental techniques developed over the past two granting periods that clearly describe how articular cartilage is able to exhibit extremely low friction coefficients even in the absence of synovial fluid, we now propose studies to determine the contribution of boundary lubricants in synovial fluid to the functional effectiveness of the lubrication mechanism in diarthrodial joints. Our proposal is timely in light of the recent resurgence of interest in proteins in synovial fluid or in the superficial zone of articular cartilage that may assist in the tribological response. To investigate our hypotheses, the proposal incorporates some novel applications of Atomic Force Microscopy (AFM), Total Internal Reflectance Fluorescence (TIRF) microscopy, and Scanning Confocal Microscopy (SCM) techniques.

Grant:	2R01AR046532-05	
Principal Investigator:	ATESHIAN, GERARD A.	PHD
Title:	Anisotropy and Nonlinearity of Cartilage Mechanics	
Institution:	COLUMBIA UNIV NEW YORK MORNINGSIDE	NEW YORK, NY
Project Period:	2000/02/01-2009/01/31	

DESCRIPTION (provided by applicant): Articular cartilage is the bearing material of diarthrodial joints. A proper understanding of the in situ response of articular cartilage under physiological loading conditions is essential in several respects. Firstly, from a basic science perspective, a knowledge of the normal mechanical function of cartilage can provide an explanation as to how cartilage can withstand the relatively harsh environment of diarthrodial joints; it is notable that, despite considerable work in the area of cartilage mechanics, the complete state of stress and strain within contacting articular cartilage layers has not been reliably determined, and consequently the physiologic loading environment of chondrocytes has not been well characterized. Secondly, proper knowledge of cartilage functional response to physiological loading can lead to a better understanding of the pathomechanical processes that might lead to cartilage degeneration and osteoarthritis through mechanical pathways. This may serve to potentially avoid or retard such degenerative processes through various clinical modalities, which properly recognize the favorable and unfavorable loading conditions of cartilage. Finally, knowledge of the loading environment of articular cartilage (such as extracellular matrix stresses and strains and interstitial fluid hydrostatic pressure) may be useful to the development of strategies to enhance tissue healing and tissue engineering of cartilage substitutes; furthermore, the functional response of such healed or engineered tissues can be correctly compared to those of normal cartilage. In this competing continuation application, we propose to build on our advances to characterize the response of articular cartilage in situ under physiological loading conditions. In a hierarchical series of progressively more complex experiments from the joint to the cellular level, our Specific Aims are to characterize (1) the change in articular thickness under normal physiological loading conditions in the human patellofemoral joint (PFJ) simultaneously with the contact stress magnitude; (2) the peak strain in the articular layer under such loading conditions, and where it occurs; (3) the peak stress in the articular layer and where it occurs; and (4) the strain environment around chondrocytes throughout the depth of the articular layer under such conditions.

Grant:	1R01EY014105-01A2	
Principal Investigator:	BACH-Y-RITA, PAUL	MD
Title:	VISION SUBSTITUTION THROUGH THE TONGUE	
Institution:	WICAB, INC.	MIDDLETON, WI
Project Period:	2004/09/15-2009/08/31	

DESCRIPTION (provided by applicant): We propose to develop and test a tongue brain-machine interface (BMI) capable of sufficiently enhancing quality of life (QoL) of blind persons to be valuable assistive technology. Investigation will be carried out along 4 interrelated lines: 1- Optimization of the technology; 2- Psychophysical investigation of the match between TVSS system and perceptual capabilities of users; 3- Investigation of changes in perceptual motor processes and capabilities of users. 4- Monitoring and analysis of usage, challenges, and opportunities the system may afford users. Studies on 1 will be carried out in Madison. Studies on 2 will be conducted at Harvard University in Boston, where the program of psychophysical assessment will address a number of aspects of visual perception with the simulators and will also assess changes that might occur in these functions with moderate level of adaptation to the device. The program will include basic testing aimed at determining the level of pattern vision provided by the system in ways similar to testing of basic visual function in observers starting with static stimuli generated by the computer, as well as full function assessments enabling the user to combined all of the flexibility and active exploration provided by head mounted camera in a simulated environment. Studies at the University of Wisconsin, Milwaukee will address areas 3& 4 to determine the way a wearable TVSS influences the actions of the wearers from very controlled motor control assessments to Orientation and Mobility (O&M) and societal participation measures. Pilot data shows that TVSS users develop new perceptual motor skills in the first few hours of training. The population of active and adventuresome adults, while least likely to experience QoL changes, will be the most demanding consumers encountering a large variety of challenges for use of the system. Ongoing evaluation of usage and problem solving will be conducted with Ecological Momentary Assessment (EMA) using cellular phones with PDA capabilities. This will allow for compliance or usage tracking, contingent sampling and purposive sampling throughout the study on each wearer, allowing the iterative process necessary for optimization of the system. The study of the capacity of motivated blind persons to extract meaningful information with frequent use of a wearable, user friendly and slightly higher resolution version of the present system will occur in parallel with the technology optimization and psychophysics.

Grant:	1R01EY015795-01	
Principal Investigator:	BAROCAS, VICTOR	PHD
Title:	Mechanical Interaction between Aqueous Humor and Iris	
Institution:	UNIVERSITY OF MINNESOTA TWIN CITIES	MINNEAPOLIS, MN
Project Period:	2004/09/30-2008/08/31	

DESCRIPTION (provided by applicant): Iris contour is critical in angle-closure, pigment dispersion syndrome, and pigmentary glaucoma. Although many studies have examined iris contour under various conditions, the basic mechanical interactions between the iris and its surroundings remain poorly elucidated. We therefore propose to perform both computational and clinical studies of iris contour under different dynamic circumstances. The word "dynamic" is emphasized as central to our hypothesis that the anterior segment is a dynamic system, and the iris contour is determined largely by dynamic effects. The effects of accommodation, blinking, and pupil diameter are not due to the position of the relevant tissues but rather to the motion thereof. For example, our hypothesis implies that the posterior bowing of the iris seen following accommodation is the result of the movement of the lens, not the final shape of the lens. Similar ramifications exist for blinking and pupil dilation and miosis, each of which is a dynamic event. The computational studies will involve extension of our previous models of the aqueous humor- iris system to include the following: dynamic active tension of the iris sphincter (already included in steady-state simulations), combines lens and iris motion during accommodation, and the dynamic mechanical effect of blinking on the anterior segment. In addition to extending our models, we will apply them to a series of periodic and random events including accommodation, blinking, and pupil diameter changes. We will compute the dynamic iris contour and will use the results to assess the mechanisms underlying angle closure and pigmentary glaucoma. The clinical studies will involve ultrasound biomicroscopy of the anterior segment during and for three minutes following accommodation. The accommodation studies will be critical because they will separate lens position (unchanged) from lens dynamics (during vs. after accommodation), We will also perform a retrospective study of dark-room angle closure tests, which will provide insight into the role of pupil dynamics in angle closure.

Grant:	1R01CA109385-01	
Principal Investigator:	BARTON, JENNIFER K	BS
Title:	Dual-Modality System for Imaging Colon Cancer in Mice	
Institution:	UNIVERSITY OF ARIZONA	TUCSON, AZ
Project Period:	2004/07/01-2009/06/30	

DESCRIPTION (provided by applicant): The overall goal of this research is to create and apply a unique dual-modality system for time-series imaging of colon cancer development in mice. Miniaturized optical coherence tomography (OCT), laser induced fluorescence (LIF), and novel contrast agents are combined into a sensitive structural and biochemical imaging system. The minimally invasive nature of this system means that the development (and response to treatment) of each individual tumor can be tracked in vivo. This study is divided into 4 specific aims: 1. Design, build and test miniature combined OCTILIF catheters. We propose to build a high-throughput probe with ability to measure the entire lower colon with one insertion. Subsequently, we propose to build a large bandwidth catheter that supports very high resolution imaging. 2. Develop novel contrast agents for improved visualization of tumors. Contrast agents may help identify the earliest stages of carcinogenesis. We will investigate three classes: a) gold-coated nanoshells, which we have shown to have an extremely bright OCT signature, b) liposomes, which incorporate fluorescent dyes, and c) dietary cholorphyll, These agents will be tested in phantoms, cell cultures, and mouse models. 3. Perform studies of DMH-treated and MIN mice with and without contrast agents and NSAID administration. Two mouse models, DMH-treated and genetically altered multiple intestinal neoplasia (MIN) will be utilized in this specific aim. In a series of studies, the lower colon of these mice will be imaged every 3-6 weeks to evaluate tumorigenesis. The effect of non-steroidal anti-inflammatory agent (NSAID) preventive and therapeutic treatment will be assessed, as will enhancement of image contrast with the contrast agents in specific aim 2. 4. Develop analysis tools for interpreting OCT images and LIF spectra. Storage and retrieval software will be developed. For analyzing OCT images, texture analysis and automated layer thickness measurements will be used, and for LIF spectra, component analysis and intrinsic fluorescence extraction methods will be employed.

Grant:	1R01HL072011-01A2	
Principal Investigator:	BEARD, DANIEL A	BS
Title:	Integrated Modeling of Cardiac Metabolism and Transport	
Institution:	UNIVERSITY OF WASHINGTON	SEATTLE, WA
Project Period:	2004/04/01-2004/04/02	

DESCRIPTION (provided by applicant): The overall goal of this proposed theoretical study is to develop cardiac tissue computer modeling tools to be used for the following applications: (1) analysis of steady-state data on myoglobin saturation and oxygen distribution in isolated perfused hearts; (2) analysis of transient data on energetics metabolism and oxygen transport in cardiac tissue; and (3) design and analysis of experiments for probing signaling and control mechanisms linking metabolic and transport processes. The proposed project takes a step toward a (more) complete multi-scale description of molecular, cellular, and tissue function by focusing on the coupling between substrate transport and cellular energetics in cardiac muscle tissue. The tools that we develop will integrate state-of-the-art knowledge of the relevant pathways of metabolism and energy transduction in working cardiomyocytes, oxygen transport in cardiac muscle tissue, and microvascular blood flow in coronary capillary networks. Spectrographic measurements of myoglobin saturation in isolated perfused guinea pig hearts have prompted us to develop a transport model for oxygen in cardiac tissue to be used to analyze and interpret the experimental findings. With this model, we are beginning to make quantitative sense of the data from the isolated hearts. Next, we aim to extend this model to link to a model of distributed energy transduction. This integrated model will allow us to investigate more complex phenomena, including transient data on oxygen uptake and 31P-NMR data on cardiac energetics. Further applications of the proposed integrated model include profiling physiological responses to several challenges, such as (1) exercise; (2) ischemia; and (3) myoglobin and adenylate kinase gene deletions.

Grant:	1R01HL075489-01A1	
Principal Investigator:	BUCKNER, GREGORY D	PHD
Title:	Innovative Tools & Techniques for Robotic Heart Surgery	
Institution:	NORTH CAROLINA STATE UNIVERSITY RALEIGH	RALEIGH, NC
Project Period:	2004/08/16-2007/07/31	

DESCRIPTION (provided by applicant): Today's surgical robots employ generalized end effectors that directly extend the motions, capabilities, and limitations of a surgeon's own hands. These robots give surgeons the ability to work on very small scales, with great precision, and through smaller incisions. They provide direct visualization through the use of magnified 3-D images and greater accuracy through motion scaling and active filtering of hand tremors. In the specific field of cardiac surgery, minimally invasive robot-assisted (MIRA) procedures show improvements in patient satisfaction and key outcome parameters including decreases in overall hospital stays. Unfortunately, these gains have been offset by significantly increased operative times, resulting in increased overall healthcare costs. Based on East Carolinas University's experience in MIRA mitral valve repairs, patient bypass times are currently increased approximately 60% (2.6 hours with MIRA vs. 1.5 hours using conventional procedures). It is evident that specific technological advancements could significantly decrease MIRA mitral valve repair times. Surgeons and researchers working in this field expect that costs will dramatically decrease as surgeons and medical device manufacturers collaboratively develop robotic tools and technologies specifically suited for MIRA cardiac procedures. Additionally, these procedures could be made less invasive with "totally-endoscopic" technologies. Accordingly, the specific aims of this multidisciplinary research program focus on developing technologies that facilitate and extend the capabilities of MIRA cardiac surgery. These include: (1) Devices for rapid and secure fixation of suture materials and prosthetic devices: specifically instruments and cartridges that provide "push-button" fixation for specific procedures (e.g. atrial closure and leaflet repair) using both existing suture materials and advanced clips and staples. (2) Endoscopic retractors to improve visualization of essential cardiac structures: endoscopically-deployable retractors that utilize the superelastic properties of Nitinol to facilitate totally closed surgical procedures. (3) Systems to aid the surgeon in incision planning, robotic navigation, and operative training: technology that can be used to measure and register critical anatomical landmarks with pre-operative and intra-operative spatial data to identify optimal port placement and robot instrument trajectories.

Grant:	1R01AI059517-01	
Principal Investigator:	CHANDLER, DARRELL P	PHD
Title:	Multiplexed pathogen detection by on-chip amplification	
Institution:	UNIVERSITY OF CHICAGO	CHICAGO, IL
Project Period:	2004/03/01-2009/02/28	

DESCRIPTION (provided by investigator): Whether in clinical or epidemiological (public health and environmental) settings, there is a continued need to detect pathogens before the onset of clinical symptoms; for Category A-C agents in particular, this need translates into de-centralized devices and methods for detecting trace organisms in large-volume environmental samples. Viability, infectivity and live/dead status of the target pathogen may be an important indicator or requirement, requiring the simultaneous analysis of DNA and RNA in the same sample. Thus, the public health pathogen detection predicament presents unique challenges to current microfluidic PCR and/or array detection devices. Solution-phase PCR by itself, however, is limited in the number of gene targets that can be accessed within a single sample and optical interference with common (TaqMan-like) reporters and molecular beacons. The objective of this application is to overcome these deficiencies and develop an integrated, 3-dimensional gel pad sample purification and amplification/detection chip to detect Category A-C pathogens in the environment. Specific aims include developing a common, high-throughput biochip platform for simultaneous, on-chip DNA and RNA purification; on-chip PCR and RT-PCR methods for ultra-sensitive detection of low-abundance nucleic acids within complex environmental samples; methods for PCR chip fabrication that can be widely disseminated and used by others, within and beyond centralized diagnostic laboratories; and a l00-plex amplification chip targeting Category A-C pathogens. We will meet these objectives by taking advantage of long-standing work in automated affinity separations for environmental samples; Argonne's unique 3-dimensional gel-pad microarrays to immobilize affinity probes in a solution-phase, spatially ordered array; new (proprietary) gel compositions that support within-gel thermal cycling and nucleic acid amplification; and on-going DoD instrument development activities for infield biochip imaging and analysis. We will validate the technology on BSL-2 bacterial pathogen DNA and RNA targets in pure culture and amended aerosol, surface water (river, marsh, pond) and soil samples. Successful demonstration of a highly multiplexed RT-PCR chip in an amended environmental sample will lay the foundation for the development of distributed diagnostic systems for the rapid detection and characterization of pathogens in the natural environment, and further validation testing of the prototype systems in physiological/clinical samples.

Grant:	1R01DK068401-01	
Principal Investigator:	CHEN, WEILIAM	PHD
Title:	Wound Repair by Stimulation of Specific Tissue Ingrowth	
Institution:	STATE UNIVERSITY NEW YORK STONY BROOK	STONY BROOK, NY
Project Period:	2004/09/01-2008/06/30	

DESCRIPTION (provided by applicant): Diabetes is the 6th leading cause of medically related death in the United States. Development of foot ulcer is one of the most serious complications, affecting up to 3% of the diabetic population each year, with 15% of all diabetic individuals experiencing one episode during their life-time. Most current treatments (e.g., off-weighting, debridement and ischemic reversal) provide only limited benefits. Recombinant human platelet derived growth factor (PDGF-BB) (becaplerrnin 0.01% gel, Regranex TM) has been approved by the FDA for topical use in treating diabetic ulcer. Compared to placebo, the incidence of complete ulcer closure was increased in some clinical trials, but not all. Non-optimal dosing was likely a major contributory factor to the discrepancy observed. We intend to engineer a biodegradable composite bioactive matrix that mimics the initial provisional matrix formed after acute wounds to achieve better healing response for chronic wounds. The initial function of this matrix is not to achieve prolonged release of bioactive agents into the wound site; rather, the therapeutic moieties will largely be confined to the matrix. This system will be constructed from Hyaluronan (HA) and a heparin stabilized cell adhesion molecule Fibronectin (FN), capable of stimulating cell migration, will be integrated into the HA structure. Both HA and FN are the main components of the provisional matrix. Additionally, heparin stabilized Vascular Endothelial Growth Factor (VEGF), with limited diffusion potential, will be incorporated to locally stimulate the endothelial cells to form blood vessels; setting the stage for granulation tissue formation. An integral component of the matrix will contain DNA encoding PDGF intended for delayed release and expression, which helps to promote the formation of robust and mature blood vessels. Migration of cells into the matrix in conjunction with blood vessel growth will precede PDGF gene transfer and subsequent PDGF expression, which will enhance granulation tissue formation. HA is truly biocompatible and does not induce inflammatory and immunogenic responses, and has mechanical properties compatible with soft tissues. Additionally, the HA matrix structure will eventually be integrated into the tissue formed during the healing process. We will first formulate and optimize these bioactive matrices using photocrosslinkable HA. The dosing information will be derived using mouse cremaster muscle models followed by a test of efficacy in diabetic mice full-thickness dermal wound models. We will use a noninvasive optical biopsy technique (Optical Coherence Tomography, OCT) to monitor the recovery of the wounds.

Grant:	1R01AR051497-01	
Principal Investigator:	CHOLEWICKI, JACEK	PHD
Title:	Mechanism and Function of Lumbosacral Orthoses	
Institution:	YALE UNIVERSITY	NEW HAVEN, CT
Project Period:	2004/09/01-2007/05/31	

DESCRIPTION (provided by applicant): New mechanisms and function of lumbosacral orthoses (LSOs) in the management of low back pain (LBP) are explored in this application by testing four hypotheses. These hypotheses are based on the rationale of LSO-enhanced spine stability, which has not been considered in this context before. The LSO provides additional stability to the lumbar spine and, therefore, patients with LBP will (i) exhibit less trunk antagonistic muscle co-contraction when wearing the LSO as compared to the "No LSO" condition, (ii) respond with a smaller number of muscles and longer latency to sudden trunk loading when wearing the LSO as compared to the "No LSO" condition, (iii) walk with gait closer resembling a normal pattern when wearing the LSO as compared to walking without the LSO, and (iv) exhibit enhanced proprioception in the lumbar spine when wearing the LSO as compared to the "No LSO" condition. Twenty LBP subjects for the experimental group and 20 control LBP subjects will be recruited from the Yale Spine Service clinics to test each hypothesis. The subjects in the experimental group will wear the LSO for 4-6 weeks. All subjects will be tested every two weeks with and without the LSO in 3 separate experiments to address each hypothesis. These experiments will consist of (1) the EMG Experiment to address hypotheses (i) and (ii), (2) the Gait Experiment to address hypothesis (iii), and (3) the Proprioception Experiment to address hypothesis (iv). New research developments will allow us to overcome several methodological and conceptual difficulties to make the proposed research now feasible. Therefore, the application of the following features will distinguish the proposed research from past studies: (i) the concept and quantification of spine stability, (ii) an experimental design tuned to detect very small LSO effects in EMG activity, (iii) a novel, submaximal exertion procedure for normalizing EMG data for patients with LBP, who are reluctant or unable to exert maximum effort, and (iv) a long-term (4-6 weeks) study, which may be necessary for acclimatization to the LSO. The results from these studies could be used in formulating rational guidelines for the prescription of LSOs as well as for improving their designs. In addition, data generated from the proposed research will also carry-over into advancing our understanding of the mechanisms and function of ergonomic lumbar supports.

Grant:	1R01EY014975-01A1	
Principal Investigator:	DE BOER, JOHANNES F	PHD
Title:	Video Rate OCT for Evaluating Glaucoma	
Institution:	MASSACHUSETTS GENERAL HOSPITAL	BOSTON, MA
Project Period:	2004/04/01-2007/03/31	

DESCRIPTION (provided by applicant): Glaucoma is the second leading cause of blindness worldwide. In the United States, approximately 2.5 million Americans are affected by this potentially blinding eye disease. Optical coherence tomography is the only non-invasive imaging technique allowing high-resolution cross-sectional imaging of the human retina. Since glaucoma causes thinning of the retinal nerve fiber layer prior to initial loss of vision, optical coherence tomograhy (which can measure the nerve fiber layer thickness) could enable early detection of glaucoma prior to any permanent loss of vision. This earlier detection would enable earlier treatment to prevent permanent loss of vision. Since glaucoma also causes progressive nerve fiber layer thinning prior to further loss of vision, optical coherence tomography could enable earlier detection of glaucomatous disease progression prior to further permanent loss of vision, enabling more aggressive preventive treatment. Current clinical techniques only allow diagnosis of glaucomatous loss of vision after up to half of the retinal ganglion cells are permanently lost. The overall goal of this research is A) to develop a video rate Optical Coherence Tomography (OCT) system for 3 dimensional high resolution imaging of the human retina and the retinal nerve fiber layer (RNFL), B) to determine the resolution and reproducibility of in vivo retinal nerve fiber layer thickness determination with video rate OCT in normal and glaucoma subjects, C) to correlate in vivo human OCT images with histology of the same eye.

Grant:	1R01EB001467-01A1	
Principal Investigator:	ESENALIEV, RINAT O	PHD
Title:	Continuous glucose monitoring in critically ill patients	
Institution:	UNIVERSITY OF TEXAS MEDICAL BR GALVESTON	GALVESTON, TX
Project Period:	2004/03/01-2007/02/28	

DESCRIPTION (provided by applicant): In both nondiabetic and diabetic patients, hyperglycemia and insulin resistance commonly complicate critical illness. Even moderate hyperglycemia, at levels that conventionally have not been treated acutely with insulin because of the risk of inducing hypoglycemia, contributes to morbidity and mortality. A recent randomized clinical trial in critically ill patients demonstrated that intensive insulin therapy to tightly control blood glucose concentration (80 - 110 mg/dL) substantially reduced morbidity and mortality by more than 40% (from 8.0% to 4.6%) but was associated with a 5.0% incidence of severe hypoglycemia (glucose concentration < 40 mg/dL). Therefore, in critically ill patients, continuous glucose monitoring, ideally noninvasive, would be invaluable to guide insulin infusion to both control hyperglycemia and avoid hypoglycemia. For this purpose, optical coherence tomography (OCT) based on low-coherence interferometry, a high-resolution optical technique that sensitively detects photons coherently scattered from tissue, is highly promising. We have developed a novel, OCT-based glucose sensor that precisely, continuously and noninvasively measures the decrease of tissue light scattering that linearly accompanies increases of blood glucose concentration. During the past two years, supported by an NIDDK R-21 grant under the PA-99-036 ("Pilot and Feasibility Program in Diabetes Endocrinology and Metabolism"), we performed preliminary animal and clinical studies of the novel glucose sensor. Our studies demonstrated: 1) a sharp and linear decrease of the OCT signal slope in skin and oral mucosa as blood glucose concentration increased; and 2) substantial improvement of accuracy of the OCT signal slope measurement by optimizing the dimensions of the probed tissue area. The goals of the proposed project are: (1) to further refine the glucose sensor in animal studies; and (2) to validate the resulting sensor in clinical studies in normal subjects and critically ill patients. Successful implementation of the project will produce a continuous, noninvasive, and accurate glucose sensor that will substantively contribute to reduced mortality and morbidity in critically ill patients.

Grant:	1R01AI056318-01A1	
Principal Investigator:	FARAG, SHERIF S	MD
Title:	QMS Technology to Deplete T Cell Alloreactivity	
Institution:	OHIO STATE UNIVERSITY	COLUMBUS, OH
Project Period:	2004/05/01-2009/04/30	

DESCRIPTION (provided by applicant): Allogeneic hematopoietic stem ceil transplantation remains the only curative option for many patients with hematological malignancies. Graft-versus-host disease (GvHD) is a major limitation to transplantation of hematopoeitic cells across histocompatibility barriers, and effectively limits transplantation to a minority of patients who would benefit from treatment. Extensive T cell depletion of the donor graft can eliminate GvHD. A significant consequence of T cell depletion, however, is a profound and long-lasting T cell immunodeficiency state post-transplant resulting in severe opportunistic viral and fungal infections that significantly limit survival. The adoptive infusion of mature, memory T cells post-transplant may offer protection against opportunistic infection, and shorted the period of immunodeficiency until successful immune reconstitution occurs. The success of this strategy in reducing mortality following T cell depleted transplants, however, will depend on the selective removal of alloreactive T cells that mediate GvHD, while retaining a high repertoire of memory T cells capable of reacting to viral and third party antigens. Although a number of methods have been investigated, the extent of depleting alloreactive T cell has been limited only to <3 log, which has not fully prevented GvHD and limited the number of T cells that can be infused to improve immunity. Either incomplete in-vitro activation of donor T cells with alloreactive potential and/or inefficient depletion of the activated cells likely contributes to suboptimal depletion of alloreactivity. The overall goal of this proposal is to develop a system for efficient depletion of alloreactive T cells for clinical use. Specifically, we aim to 1) Optimize the conditions for activating the maximum number of donor-specific T cells by studying culture conditions and the kinetics of different antigens expressed selectively on activated T cells for use as targets in depletion, and 2) Develop a GMP grade hiqh-performance immunomagnetic separation system, quadrupole magnetic cells sorting (QMS), for clinical scale depletion of activated donor T cells that is capable of >=3 log depletion of alloreactive T cells while retaininq >80% of third party reactivity. The technology developed in this proposal should greatly facilitate the development of clinical trials of adoptive donor T cell therapy to improve immune function following T-cell-depleted mismatched stem cell transplants.

Grant:	1R01CA098465-01A2	
Principal Investigator:	FELEPPA, ERNEST J	
Title:	Novel Ultrasonic Imaging of Brachytherapy Seeds	
Institution:	RIVERSIDE RESEARCH INSTITUTE	NEW YORK, NY
Project Period:	2004/08/03-2008/07/31	

DESCRIPTION (provided by applicant): Prostate brachytherapy, which implants radioactive seeds throughout the gland in order to kill cancerous tissue, is an increasingly popular method of treating localized prostate cancer. In the US, the number of implants increases by about 10% annually, and an estimated 50,000 to 60,000 procedures were performed in 2002. The five-year survival rate for prostate brachytherapy is comparable to the survival rate for radical prostatectomy. The placement of seeds is planned either in advance of treatment or in the operating room using dosimetry software that incorporates conventional ultrasonic images of the prostate and its surrounding anatomy. However, seeds often become displaced because of movement and distortion of the gland during seed insertion, and conventional ultrasonic imaging does not effectively show the actual location of implanted seeds. Consequently, some targeted regions may receive an insufficient dose. As a result, at least 15,000 procedures in the US provided inadequate dosage in 2002. An improved means of visualizing seeds would enable corrective seed placements to be performed and could result in marked improvement in treatment efficacy. The research proposed in this application seeks to develop novel advanced ultrasonic imaging methods that overcome the limitations of conventional ultrasonic instruments now used for imaging the prostate and implanted seeds. The proposed studies will investigate correlation, resonance, modified-elastographic methods applied alone and in combination, using tissue-mimicking phantoms, in vitro tissue specimens, animals and human subjects. If successful, the study will result in improved sensitivity and specificity in ultrasonic images of seeds. The proposed research will be performed collaboratively by engineers and scientists at Riverside Research Institute working with medical physicists and radiation oncologists at the New York Presbyterian Medical Center of Columbia University. Technical support in the form of consulting, software, seeds, etc., will be provided by Varian Medical Systems, Inc., which offers a potential path for translating the research results into a commercial product.

Grant:	1R01NS046602-01A1	
Principal Investigator:	FREI, MARK G	PHD
Title:	Synchronism and nonliner dynamics in seizure prediction	
Institution:	FLINT HILLS SCIENTIFIC, LLC	LAWRENCE, KS
Project Period:	2004/08/01-2007/06/30	

DESCRIPTION (provided by applicant): Seizure prediction and blockage of seizures is a top research priority in epilepsy, a disease affecting about 1% of the US population. Due to the extreme complexity of brain dynamics, its analysis requires sophisticated methodologies and algorithms from applied mathematics, physics, and engineering. Much emphasis has been placed on techniques from nonlinear dynamics, including application of synchronization and dynamical invariants techniques. In this proposal, we will perform a systematic assessment of these techniques for the task of seizure detection and prediction on long time series. First, we look for optimal synchronization measures, examining phase synchronization, generalized synchronization, and unstable periodic orbit analysis, and assessing each for its sensitivity and specificity for prediction or detection of epileptic seizures. We will test these strategies as well Lyapunov exponents on multi-day, continuous ECoG recordings from several patients, with each recording containing several seizures.

Grant:	1R01HL075515-01	
Principal Investigator:	GILMOUR, ROBERT F	
Title:	Computer Model of the Canine Ventricle	
Institution:	CORNELL UNIVERSITY ITHACA	ITHACA, NY
Project Period:	2003/12/15-2007/11/30	

DESCRIPTION (provided by applicant): Sudden death secondary to ventricular fibrillation (VF) remains a leading cause of mortality in the US. Therapy for VF has been largely ineffectual, principally because the underlying mechanisms for VF are not well understood and probing for potential mechanisms has been hindered by the inability to precisely modify specific ionic currents. To address these issues, we propose to develop a data-driven computer model of the electrical behavior of the canine ventricle. Specifically, we will: 1) Experimentally characterize IKr, ICa, IK1, INaCa, and the late sodium current INa in myocytes obtained from specific regions of the ventricles. These particular currents will be studied because they play a significant role in repolarization. They will be measured using action potentials recorded at rapid pacing rates as the command waveforms, to replicate current behavior during a tachyarrhythmias. 2) Develop deterministic Hodgkin-Huxley and Markov models for each ionic current for each anatomical region using the time series and steady state current data obtained under Specific Aim 1. Optimization routines will be used to determine unknown parameters in the models by comparing the model current to experimental data. 3) Incorporate the models of the individual currents into computer models of region-specific single canine ventricular myocytes. Models of left and right ventricular epicardial, midmyocardial and endocardial myocytes of basal and apical origin and of right and left ventricular Purkinje myocytes will be developed. 4) Incorporate the single cell models into a 3-D computer model of the canine ventricle using a modified version of the phase field method. The model will be written using a portable parallel version of the code and run on a parallel computer and multi-node clusters. Initially, the model will consist of the left ventricle, with epicardial, midmyocardial and endocardial layers. More detailed anatomical models subsequently will be constructed to include the His-Purkinje system and the right ventricle. 5) Use the 3-D model to test candidate hypotheses for the development of VF. The initial test will determine whether suppressing dynamic electrical heterogeneity prevents VF. The computer model of ventricular electrical function we propose will provide an invaluable tool for drug discovery and the evaluation of algorithms for anti-tachycardia and anti-fibrillatory pacing and defibrillation. As such, the model is expected to have a significant impact on the diagnosis and treatment of lethal heart rhythm disorders.

Grant:	1R01AR051336-01	
Principal Investigator:	GULDBERG, ROBERT E	PHD
Title:	Vascularization and Bone Repair	
Institution:	GEORGIA INSTITUTE OF TECHNOLOGY	ATLANTA, GA
Project Period:	2004/07/01-2008/06/30	

DESCRIPTION (provided by applicant): The requirements for bone repair, whether provided by the host or within an implantable tissue-engineered construct, include an extracellular matrix scaffold, cells, a functional vascular supply, and osteoinductive factors. Of these essential elements, vascular supply has been the least studied in the context of tissue engineering in part due to the difficulty in quantifying 3-D vascular structures within tissues. Traditionally, 2-D histological analysis has been used to assess vessel density. However, this approach is semi-quantitative at best and does not easily allow analysis throughout the tissue. High-resolution 3-D microcomputed tomography (micro-CT) imaging coupled with contrast agent perfusion has the potential to overcome these limitations to quantify vascular growth. The recent development of in vivo micro-CT systems has further provided the opportunity to non-invasively monitor mineralized matrix formation within a bone defect in vivo. The goal of this application is to combine and extend these methodologies to better understand the temporal and spatial relationships between vascularization and mineralization in a well defined in vivo bone tissue engineering model. The Specific Aims are to: I. Quantify 3-D vascular growth and mineralized matrix formation within scaffolds implanted into critically-sized segmental bone defects, II. Analyze the influence of porous scaffold architecture on vascular invasion and mineralization, and III. Test the effect of adding a cellular component to implanted constructs on vascularization and mineralization during segmental defect repair. The proposed research is highly significant because it integrates quantitative 3-D imaging techniques with a well characterized in vivo model to better understand the inter-relationship between two processes essential to bone repair: vascularization and mineralization. Lack of a vigorous vascular response may be an important mechanism of delayed or failed bone repair. Decoupling of vascularization and mineralization responses is also possible during for example fibrous tissue repair. The restoration of a functional vascular supply is a critical issue for engineering the repair of a wide variety of tissues in addition to bone. Thus, the proposed studies will establish a valuable new approach for assessing the integration of tissue-engineered constructs into living systems. Furthermore, the developed methodologies will have broad applicability to other research areas, including for example studies on fracture healing, skeletal development, vascular injury, and tumorigenesis.

Grant:	1R01HL071958-01A1	
Principal Investigator:	HELMKE, BRIAN P	BS
Title:	Flow-Induced Cytoskeletal Mechanics in Endothelial Cells	
Institution:	UNIVERSITY OF VIRGINIA CHARLOTTESVILLE	CHARLOTTESVILLE, VA
Project Period:	2004/01/01-2007/12/31	

DESCRIPTION (provided by applicant): Endothelial cell (EC) adaptation to the complex local hemodynamic environment plays a critical role in both the physiological and pathological regulation of vessel wall biology, but mechanisms by which ECs transduce fluid mechanical forces into biochemical signals remains poorly understood. This proposal will address two key questions related to the initiation of mechanotransduction: is extracellular applied fluid force transmitted to the interior of the cell where biochemical signaling molecules are located, and is flow-induced intracellular deformation concentrated at discrete locations in the cell at a magnitude that mediates structural protein interactions? High-resolution 4-D microscopy imaging of green fluorescent protein fused to vimentin, actin, and paxillin will enable measurements to test the hypothesis that changes in extracellular applied fluid shear stress induce spatially focused mechanical responses in the cytoskeleton near intracellular locations where structural proteins are involved in rapid mechanochemical signal transduction. This hypothesis suggests that strain focusing by local cytoskeletal deformation provides the spatial organization of structural proteins necessary to trigger specific biochemical signaling networks in response to changes in the hemodynamic environment. The specific aims are (1) to determine the spatiotemporal distribution of strain focusing in the actin microfilament network in living ECs during a change in shear stress, (2) to determine the relative contributions of microfilament and intermediate filament networks in focusing cytoskeletal strain during onset of shear stress, and (3) to determine whether focusing of cytoskeletal strain in response to shear stress occurs near sites of focal adhesion to the extracellular matrix and initiates spatial redistribution of focal adhesion proteins. Since focal adhesion proteins are rapidly phosphorylated by onset of shear stress, signaling at these locations may be initiated by mechanical interactions with the cytoskeleton. A novel measurement of interaction strain will be defined to indicate the degree of structural rigidity of mechanical connections between the cytoskeleton and focal adhesion sites. This proposal will measure for the first time spatial and temporal relationships between mechanical interactions in the cytoskeleton and locations involved in initiation of mechanotransduction. The long-term goal of this research program is to define biomechanical mechanisms contributing to cell and tissue function in order to develop innovative approaches for treating endothelial dysfunction in vascular pathology and artificial graft design.

Grant:	1R01HL077606-01	
Principal Investigator:	HYNYNEN, KULLERVO H	
Title:	Phased Array Ultrasound System for Cardiac Ablation	
Institution:	BRIGHAM AND WOMEN'S HOSPITAL	BOSTON, MA
Project Period:	2004/07/06-2007/06/30	

DESCRIPTION (provided by applicant): Cardiologic intervention for the treatment of tachyarrhythrnias has evolved from pharmacological therapy to surgical based elimination of arrhythmogenic loci and circuits to catheter based ablation procedures. Ablation via percutaneously placed catheters has become the standard form of therapy for many tachyarrhythmias, surpassing pharmacologic and surgical methods. Despite these major advances, catheter ablation has major limitations effecting both efficacy and safety. The inability to accurately identify anatomic targets and create transmural, continuous lesions limits efficacy. Many of the complications associated with this procedure are due to its invasive nature. In addition, the procedure is performed under fluoroscopy guidance that during a prolonged procedure exposes the patient and physician to a significant dose of ionizing radiation. In this research we will test a hypothesis that an ultrasound phased array with full three dimensional beam steering capability placed in the esophagus and guided and monitored by magnetic resonance imaging can induce accurately targeted transmural myocardial lesions without the problems associated with the intracardiac catheter placement. To accomplish this we plan to: First, perform in vivo animal tests with our linear phased arrays to explore the sonication parameters and methods for inducing adequate tissue coagulation. Second, perform a computer simulation study to explore the array design and sonication methods for trans-esophageal cardiac ablation. Third, develop and test the phased array applicators both in ex vivo and in vivo animal tissues, and fourth, develop MR thermometry methods that can monitor and guide cardiac muscle coagulation. Finally, we plan to develop a complete sonication system and test it in animal experiments. The ability to image in real time the true anatomy without exposure to ionizing radiation and to precisely target and create transmural lesions non-invasively would be a major revolution in the treatment of cardiac arrhythmias. This could result in significant cost savings and lead to a cure of thousands of patients.

Grant:	1R01NS048903-01	
Principal Investigator:	KACZMAREK, KURT A	PHD
Title:	Electrotactile Sensory Magnitude and Quality	
Institution:	UNIVERSITY OF WISCONSIN MADISON	MADISON, WI
Project Period:	2004/07/01-2008/06/30	

DESCRIPTION (provided by applicant): Electrical stimulation of touch (electrotactile or electrocutaneous stimulation) has shown promise for a wide variety of applications in which information is presented tactually to human users. These include sensory prostheses for persons with serious visual and auditory impairments, as well as for those who have lost tactile sensation on some cutaneous loci due to traumatic nerve injury or disease, and also for advanced robotic surgical techniques and other applications. Over the last thirty years, many such concepts been implemented and tested. However, these efforts have been largely application-specific and in general hampered by a lack of both theoretical framework and fundamental research aimed at understanding the basic perceptual characteristics of the electrotactile response. The proposed discovery-driven research aims to study the intensive and qualitative psychophysics of this unique method of information display, and specifically to: (1) Characterize the dynamic range of the electrotactile percept as a function of physical stimulus parameters, particularly those (such as cutaneous locus and hydration) that historically have made it difficult to control the electrotactile percept, and (2) Characterize the qualitative or subjective nature of the electrotactile percept ("tactile color") as a function of waveform timing. These results will enable our future goal of developing specific stimulus control schemes and stimulus parameters that maximize the dynamic range and controllability of the electrotactile percept. The long-term result will be superior tactile communication means for medical, industrial, and consumer applications. A secondary result will be a preliminary but quantitative description of the electrotactile stimulus-percept space; this will aid future research aiming to uncover the physiological mechanisms underlying electrical stimulation of touch.

Grant:	2R01EB002091-04	
Principal Investigator:	KILGORE, KEVIN L	PHD
Title:	A Novel Waveform for Electrical Nerve Conduction Block	
Institution:	CASE WESTERN RESERVE UNIVERSITY	CLEVELAND, OH
Project Period:	2001/06/15-2007/04/30	

DESCRIPTION (provided by applicant): An effective, quick-acting and quick-reversing means of blocking the conduction of action potentials in whole nerve would have many important clinical applications, such as the elimination of pain and the modulation of muscle spasticity. One potential method for achieving this type of nerve conduction block is to use high frequency alternating current delivered through electrodes surrounding the target nerve. In our currently funded project, we have determined that this method can block 100% of the motor response in motor nerves. The block is gradable and can be completely reversed within one second. We are now ready to take the next step toward practical clinical implementation of this technique. Therefore, the specific aims of the proposed project are to: 1) determine if the chronic block of nerve conduction is damaging to the nerve, 2) determine the specific parameters necessary to provide motor and sensory block in humans, and 3) complete the technical and regulatory effort necessary to proceed to widespread human application of this technique. Chronic nerve block will be applied to the peripheral nerves of dogs for four weeks using parameters and regimens based on anticipated human applications. The post-operative nerve histology will be the primary outcome measure that will be used to determine if there are any damaging effects of the chronically applied conduction block in-vivo. We expect that the conduction block will not damage peripheral nerve because it uses a very low charge delivery per phase and delivers a zero net charge to the tissue. In conjunction with the chronic animal study, we will commence acute human studies using: 1) explanted human nerves to determine the parameters necessary for conduction block in large diameter nerves, 2) acute intra-operative testing to determine if successful motor block can be accomplished in the presence of intact reflex circuits, and 3) short-term chronic experiments to determine the parameters necessary for complete pain block. At the end of the proposed three year project, we expect to have completed all of the necessary tasks to enable the application of electrical nerve conduction block in humans for the elimination of peripheral nerve pain and for the modulation of muscle spasticity. This research will provide a significant tool in our ongoing efforts to reduce disability and improve quality of life.

Grant:	1R01AR051582-01	
Principal Investigator:	KLEIN, TERI E	PHD
Title:	Linking Collagen Genotypes to Molecular Phenotypes	
Institution:	STANFORD UNIVERSITY	STANFORD, CA
Project Period:	2004/08/05-2008/06/30	

DESCRIPTION (provided by applicant): Collagen is the most abundant protein in humans, and mutations in collagen can lead to death or disease. The ways in which individual mutations affect collagen structure/function are poorly understood. Collagen is a fibrillar protein, and thus many of the principles elucidated for the study of globular proteins are not immediately applicable in investigating the relationship between its structure and function. We now have an opportunity to use genomic technologies to survey the variation in key collagen genes throughout the human population, link the discovered polymorphisms to their structural effects, and develop an understanding of the mechanism of collagenous disorders. This work will also provide a foundation for engineering new treatments and the designing of novel collagen-like biomaterials. The long-term goal of this proposal is to determine the chemical, physical and structural properties of biopolymers in the context of natural sequence variation. We will take advantage of our joint capabilities in genomics and structural biology to pursue the following specific aims: (1) to identify Single Nucleotide Polymorphisms (SNPs) in an ethnically diverse population to determine the background genetic variation across the collagen genes COL1A1, COL1A2, COL2A1 and COL3A1 and then determine the distribution of SNPs in individuals with collagen disorders; (2) to model collagen-like peptides and full-length Type I and Type III collagen triple helices to determine the structural and energetic effects resulting from single point glycine substitutions in the collagen-like peptides and genetic variation in the full-length collagen models.; (3) to develop analysis tools that incorporate function and phenotypes observed in molecular dynamics simulations to construct a working model of type I collagen for OI-associated mutations; and, (4) to develop methods to simulate the folding and unfolding of native and mutant collagen-like peptides to determine the effects induced by mutations with phenotypic consequences.

Grant:	2R21AI049541-04	
Principal Investigator:	LARSON, RONALD G	PHD
Title:	Microfabricated Device for Rapid, Portable, Viral Genome	
Institution:	UNIVERSITY OF MICHIGAN AT ANN ARBOR	ANN ARBOR, MI
Project Period:	2001/04/01-2005/08/31	

DESCRIPTION (provided by applicant): The constant threats posed by Influenza and other RNA viruses of pandemic due to antigenic shift or transfer from an animal host, and of season-to-season variation due to antigenic drift can be most effectively countered by pervasive monitoring. Viral monitoring in the clinic and field would be assisted greatly by the availability of microfabricated devices capable of rapid, inexpensive genotyping. To accelerate the emergence of such devices we propose research to engineer and build two prototype microfabricated devices, one to detect influenza A sub-types, and the other to perform single nucleotide polymorphism (SNP) detection to monitor antigenic drift. We will also develop and integrate a purification system to prepare sufficient quantity and purity of RNA material from virus-containing clinical samples for the microfabricated device to perform reverse transcription-PCR (RT-PCR) and genotyping reactions. Specifically, we aim to Aim 1 - Develop a microfabricated device capable of producing viral RNA from biological samples with the levels of purity and concentration required to perform on-chip RT-PCR reactions. Aim 2 - On a microfabricated device, perform an RT-PCR reaction on HA 1 hemagglutinin domain of influenza A, producing double-stranded complementary DNA. Aim 3 - On a microfabricated device, perform restriction digestion reactions that can distinguish H1, H3, and H5 types of influenza A. Perform a multiplex reaction that can simultaneously detect RNA from influenza A and another RNA virus, such as influenza B or SARS. Aim 4 - On a microfabricated device, perform single-nucleotide polymorphisms that can distinguish antigenic drift in the HA1 domain of influenza A. Aim 5 - Integrate RT-PCR, restriction digestion, and electrophoretic separation onto a single microfabricated device, and integrate this with a system for RNA extraction from clinical, throat-culture, samples.

Grant:	1R01EB004108-01	
Principal Investigator:	LASKARIS, EVANGELOS T	PHD
Title:	Low-Cost High-Temperature Superconducting MRI System	
Institution:	GENERAL ELECTRIC CO CORPORATE R&D CTR	NISKAYUNA, NY
Project Period:	2004/09/30-2008/08/31	

DESCRIPTION (provided by applicant): The expense involved in installing and providing cryogen service to current superconducting MRI systems has limited their widespread distribution, especially in less populous regions of the country, as well as in underdeveloped countries. The MRI scanners found in these areas are most often of the low field (0.2-0.35T) type using Permanent Magnet (PM) technology. These systems have a lower image quality and are not cost-effective when compared with superconducting systems. The objective of this program is to develop enabling technologies for a low-cost, easily siteable superconducting MRI system with a magnetic field strength and image quality comparable to existing solenoidal superconducting systems of 1.0 - 1.5 T magnetic field strength. The system will be easily siteable in underdeveloped areas since it will not require liquid cryogens and will be designed to survive power outages with minimal operator intervention. The program will: 1) Develop key magnet technologies for cryogen-free operation and power-outage survivability. 2) Develop key technologies for PITS wire including a joining method. 3) Prove the developed technologies by building an orthopedic imaging system. 4) Evaluate the performance of the orthopedic imaging system in terms of image quality and system reliability.

Grant:	1R01NS045753-01A1	
Principal Investigator:	LIEBER, BARUCH B	
Title:	Flow Divertors to Cure Cerebral Aneurysms	
Institution:	UNIVERSITY OF MIAMI CORAL GABLES	CORAL GABLES, FL
Project Period:	2003/12/01-2007/11/30	

DESCRIPTION (provided by applicant): Stroke is the most common life-threatening neurological disease and the third leading cause of death in the United States One fourth of the deaths from cerebrovascular disease in the United States arise from hemorrhage and stroke associated with rupture of intracranial aneurysms. The endovascular treatment for intracranial aneurysms that is gradually replacing conventional surgery has focused on the intra-aneurysmal deposition of occlusive materials with no regard for the pathology of the disease our hypothesis is that the hemodynamics in the parent artery/aneurysm complex can be altered by the minimally invasive implantation of a flow divertor in the parent vessel. Scaffolding by the divertor initiates parent artery/aneurysm remodeling, leading to a cure of the lesion. The design parameters, i.e., axial and radial distensibility, filament diameter, pore size, and the material composition of the flow divertor will have to be tailored to the local hemodynamics for a permanent occlusion of the aneurysm, yet preventing vessel injury, acute thrombosis, or delayed stenosis inside the bioimplant. We propose to identify the optimal design of a flow divertor for endovascular bypass of intracranial aneurysms through two sequential but complementary approaches. In the first approach, vascular replicas of experimental aneurysm models in rabbit will be used in vitro to optimize the interaction between the flow divertor and the vascular hemodynamics. Thereafter, the most promising designs will be selected for implantation in an aneurysm model in rabbit to elucidate the remodeling of the vasculature in response to the implanted flow divertor. High spatio-temporal resolution data acquired from the bench top experiments will be correlated with the limited amount of data that can be obtained in vivo. The postmortem histopathological analysis of the in vivo data will provide definitive conclusions to observations made angiographically in vivo. The tasks will be accomplished through the following specific aims: 1) To construct elastomer replicas of the rabbit aneurysm model for bench top investigation using a mock circulation loop; 2) To evaluate the influence of the design parameters of the flow divertor on intra-aneurysmal flow in the elastomer replicas; 3(a) To construct elastase-induced bifurcation aneurysm model in rabbit; (b) To implant the optimized flow divertors in the rabbit aneurysm model and quantify indices of local hemodynamic changes by the divertor; and 4) To evaluate the efficacy of the optimized flow divertors in parent artery remodeling and aneurysm exclusion.

Grant:	1R01HG003329-01	
Principal Investigator:	MATHIES, RICHARD A	PHD
Title:	Miniaturized Integrated Genotyping Microsystems	
Institution:	UNIVERSITY OF CALIFORNIA BERKELEY	BERKELEY, CA
Project Period:	2004/09/01-2009/04/30	

DESCRIPTION (provided by applicant): The objective of this grant is to develop novel microfabricated genetic analysis microsystems and associated methods that can be used for high-performance analysis of cancer genotypes in the research, discovery and/or diagnostic settings. Initial work will focus on the refinement of the apparatus, reagents and methods needed to apply Polymorphism Ratio Sequencing (PRS) to the high-throughput genetic analysis of mitochondrial DNA variations in tumor tissue using conventional capillary array electrophoresis. We will optimize the labeling and pooling methods and develop convenient PRS data analysis software. Then, Johns Hopkins University (JHU) scientists will be trained to perform PRS at UCB. Finally, we will transition the technique to JHU for its routine high-throughput application. Second, we will design, construct and evaluate a fully integrated mitochondrial PRS chip. This wafer scale device takes RCA (rolling circle amplification) prepared mitochondrial DNA and parses the template into 96 individual DNA sequencing modules, including extension reactors and CE separation channels, to produce an entire mitochondrial PRS analysis in under 1 hr. Once this system is developed, a second-generation version will be constructed and then used at JHU for high-throughput analyses. Third, we will develop a fully integrated microdevice to perform SNP and other genetic typing from genomic DNA. This device will accept purified genomic DNA as the input and will parse the individual sample to 96 different PCR reactors for multiplex allele specific amplification and analysis of polymorphisms or cancer markers. This microdevice will permit genetic typing from small quantities of DNA and has the advantage of fully integrating a large portion of the important sample preparation process thereby providing low-cost, high-throughput genotyping of tumor samples. Finally, we will develop a portable genotyping device for real-time analysis of informative mitochondiral or genomic DNA variations or diagnostic markers. This system will be valuable (i) for point-of-care genetic analysis to identify the presence of cancer markers and/or to monitor possible recurrence and (ii) for performing real-time molecular pathology of tissue samples to determine the extent of cancer invasion.

Grant:	1R01GM068587-01A1	
Principal Investigator:	MRKSICH, MILAN	BS CHEMISRY
Title:	Smart Substrates for Cell Biology and Tissue Engineering	
Institution:	UNIVERSITY OF CHICAGO	CHICAGO, IL
Project Period:	2004/09/01-2007/08/31	

DESCRIPTION (provided by applicant): This research program will develop a broad class of active substrates for mechanistic studies in cell biology and applications in tissue engineering. Most mammalian cells are adherent and must remain attached to the protein extracellular matrix in order to survive, proliferate, differentiate and function. The interactions of cells with the protein matrix are mediated by a host of cell-surface receptors and matrix-derived ligands, which serve not only to localize cells in tissue but also to provide cells with regulatory cues. In many instances, the cues are dynamically modulated: that is, the composition of ligands presented from the matrix changes over time. Experimental studies of dynamic interactions between cell and matrix are difficult and still in need of new tools that can mimic the complex patterns of interactions that are central to cellular regulation. This research program will develop a strategy for creating dynamic substrates wherein the activities of immobilized ligands can be switched in real-time under an electrical control. The program will develop model substrates wherein: i) the activities of ligands can be switched on; ii) the activities of ligands can be switched off; iii) the activities of ligands can be reversibly switched between high and low affinity states. The program will also develop multifunctional substrates that incorporate two or more of these properties. The program will apply these active substrates to a set of model problems in cell biology and tissue engineering in order to validate these strategies and meet the program goals of advancing an approach that will be valuable to experimental studies in cell biology and technologies for engineering tissue. Finally, the program will develop strategies for transitioning these active substrates to users in the biological and bioengineering communities. Classes of substrates that can be modified with a variety of ligands and that incorporate the chemistries for dynamic control over ligand activity will be developed and validated for use. This program will advance a technology that will have broad impact across several areas in biology and medicine.

Grant:	1R01DC006435-01A1	
Principal Investigator:	NAGARAJAN, SRIKANTAN S	PHD
Title:	Neural mechanisms of auditory feedback during speech	
Institution:	UNIVERSITY OF CALIFORNIA SAN FRANCISCO	SAN FRANCISCO, CA
Project Period:	2004/07/15-2009/06/30	

DESCRIPTION (provided by applicant): Understanding how auditory feedback is processed during speaking provides insights into fundamental mechanisms underlying speech production and perception. This knowledge might also ultimately contribute to the early detection and lead to treatment strategies for a number of prevalent clinical conditions where impairments in abnormal processing of auditory feedback have been reported (e.g. stuttering, Parkinson's disease, schizophrenia). While many behavioral studies have examined how auditory perception affects speech production, only recently have functional neuroimaging studies begun examining how producing speech affects the neural processes serving auditory perception. Recent studies have shown that in auditory cortex and other areas in the superior temporal plane, speaking causes "speaking-induced suppression" (SIS): response to self-produced speech is suppressed when compared to identical speech from an external source. In our recent work, we have shown that SIS in auditory cortex does not result from overall inhibition of this area during speaking. Rather, SIS appears to be a neural correlate of a feedback prediction error (FPE) - a comparison between actual auditory input and an internal "speaking-induced prediction" (SIP) of that auditory input. SIS expression in auditory cortex has led to the hypothesis that SIS reflects auditory discrimination of self-produced from externally produced stimuli (Self-non-Self Hypothesis). However, refinements in our understanding of auditory feedback in speech motor control, that are supported by behavioral studies and our preliminary data, suggest that SIS may also reflect feedback processing for speech motor control (Speech Motor Control Hypothesis). We have developed a unifying conceptual model that embodies both hypotheses, and our proposed experiments use SIS to test the neural correlates and the validity of this model. The specific aims are to determine how SIS is modulated by 1) altered feedback, 2) speech target dynamics and 3) speech motor adaptation. These manipulations not only help us to unravel the functional significance of SIS but also help us determine if there is a differentiation of the function of SIS across the superior temporal plane. Furthermore, how activity in other parts of the brain is affected by our experimental manipulations will allow us to determine the neural correlates of the mechanisms that generate SIS. Our approach capitalizes on unique real-time speech feedback alteration methods used with functional magnetic resonance imaging (fMRI) and magnetic source imaging (MSI). The excellent spatial resolution of fMRI will enable reconstruction of spatial locations of activity related to SIS and SIP while the excellent temporal resolution of MSI will enable us to reconstruct the sequence of activation in these areas.

Grant:	1R01EB000456-01A2
Principal Investigator:	NEWELL, JONATHAN C.
Title:	Breast Cancer Diagnosis By Electrical Impedence Imaging
Institution:	RENSSELAER POLYTECHNIC INSTITUTE TROY, NY
Project Period:	2004/08/02-2007/07/31

DESCRIPTION (provided by applicant): The long-term goal of this project is to develop a new technology to improve the screening for and diagnosis of breast cancer. Non-invasive electrical impedance measurements made with a hand-held robe have been shown to improve the specificity and sensitivity of mammography for breast tumor diagnosis in patients with ambiguous mammograms. This non-invasive technology poses no known risks to the subject, and provides a new diagnostic parameter to assess suspicious anomalies. This new technology, called Electrical Impedance Tomography, makes images of the interior of the body From measurements made at its surface. Small electric currents are passed through the body using electrodes applied to the skin; the resulting voltages are then measured and used by mathematical algorithms to reconstruct the values of the electrical conductivity and permittivity of the underlying structures. The electrical properties of breast tumor tissue differ by a factor of 5 -10 from those of surrounding, normal tissue. Benign breast lesions are also electrically different from malignancies. By Forming images of the electrical conductivity of the breast, tumors may be detected and differentiated. When this is done using several different frequencies of electrical current, and by measuring both conductivity and permittivity, the technique is called tissue spectroscopy, and further diagnostic information may be elucidated. We propose to determine the feasibility of improving the diagnostic ability of mammography by combining a mammography system with an adaptive current tomography (ACT) system in order to make simultaneous, in-registration images of breast electrical properties using data collected from two arrays of radiolucent electrodes applied to the mammogram plates. This is a proposal to build the required electronic instruments and electrode arrays, and to write improved algorithms to reconstruct conductivity and permittivity images from the resulting voltages and currents. It will operate at frequencies between 300 Hz and 1 MHz, using 64 electrodes, and make and display 20 frames/sec. The system will be designed for clinical use, and will be able to assess the pulsatility of blood volume, which may indicate malignancy. It will be applied in a study of normal human subjects, to determine basic operational procedures for its use. It will then be used in a clinical examination of patients undergoing biopsy for breast cancer diagnosis. Images will be made simultaneously and in register by mammography and by electrical impedance tomography. Comparison of both images with biopsy results will be made. The use of impedance images and spectroscopy for the diagnosis of breast cancer may then be able to be responsibly assessed.

Grant:	1R01HL073198-01A2	
Principal Investigator:	PINSKY, MICHAEL R	
Title:	Quantifying Left Ventricular Ejection Effectiveness	
Institution:	UNIVERSITY OF PITTSBURGH AT PITTSBURGH	PITTSBURGH, PA
Project Period:	2004/08/15-2008/07/31	

DESCRIPTION (provided by applicant): Asynchronous left ventricular (LV) contraction is the most common cardiac abnormality and, if severe, impairs LV pump function, induces cardiac dilation and heart failure remodeling. Ventricular pacing usually increases contraction asynchrony and induces cardiac dilation even when contractility is normal. We hypothesize that LV contraction asynchrony reduces LV ejection efficiency, defined by the ratio of LV stroke work to myocardial O2 consumption (MVO2), by causing LV dilation without altering intrinsic contractility. We define LV ejection effectiveness as the synchrony of contraction of all contractile elements. Importantly, recent clinical trials of cardiac resynchronization therapy (CRT) in patients with dilated cardiomyopathy and prolonged QRS have shown that gated bi-ventricular pacing improves LV ejection pressure, decreases cardiac volumes and induces reverse remodeling in some but not all subjects. We hypothesize that all the beneficial effects of CRT come from its ability to improve LV contraction synchrony. We believe that these clinically opposite effects of pacing are explained by opposite changes in contraction synchrony. The relation between MVO2, LV ejection asynchrony and ejection effectiveness is unknown. We will develop a novel application of the assessment of LV ejection efficiency combining regional phase angle analysis with Fourier analysis of both phase angle and amplitude dispersion from echocardiographic data. We propose to quantify this asynchrony at the bedside in both animal and human models using tissue Doppler imaging (TDI). We have recently developed and validated a quantitative model to assess LV ejection effectiveness using regional phase angle analysis. However, this technique requires invasive monitoring and are not suitable for general clinical use. Importantly, we have also developed and validated quantitative methods of analyzing transthoracic echocardiographic LV images using TDI and acoustic quantification (AQ) algorithms. These powerful non-invasive tools allow us to define regional myocardial movement. Presently, there is no established method of analyzing these data to objectively quantify contraction asynchrony. We propose to couple our asynchrony analysis with our quantitative AQ and TDI techniques to create a clinically relevant tool to assess LV ejection effectiveness. We will use our established isolated perfused rabbit heart (Langendorf preparation) model to validate the relation between MVO2 and asynchronous LV contraction. We will use our intact anesthetized canine model under conditions of varying contraction asynchrony induced by selective pacing, mock CRT and regional ischemia and reperfusion to create an on-line TDI analysis algorithm. Finally, we shall study human subjects before and after CRT and non-CRT subjects to ascertain if we can predict which subjects will benefit from CRT and where in the ventricle CRT pacing would be optimal. Potentially, CRT could be used in subjects before they develop heart failure remodeling. We will test two related hypotheses. First, that increased global LV asynchrony induces parallel shifts in LV volume for a constant ejection pressure such that MVO2 increases as a function of the parallel shift of the LV end-systolic pressure-volume relation. Second, that LV ejection effectiveness, measured by AQ and TDI in both clinically relevant canine models of LV contraction asynchrony and humans with cardiac disease, can be quantified as both the sum of the amplitude-corrected phase angle dispersion among LV regions and as the cross correlation of amplitude-corrected phase angles. The ultimate goal of this proposal is to develop and validate an echocardiographic-based algorithm that quantifies LV ejection effectiveness by merging both power and synchrony of contraction into a common metric.

Grant:	1R01DK064775-01A1	
Principal Investigator:	PULLAN, ANDREW J	PHD
Title:	Realistic models of gastrointestinal bioelectromagnetism	
Institution:	VANDERBILT UNIVERSITY	NASHVILLE, TN
Project Period:	2004/06/01-2009/05/31	

DESCRIPTION (provided by applicant): As reductionist biomedical science succeeds in elucidating ever more detail at the molecular level, a mathematical modeling framework will become increasingly important to cope with this explosion of data and to relate integrated whole organ function to underlying biophysically detailed mechanisms that exploit this molecular knowledge. The proposed research has two primary long term objectives. The first is to develop an extensible anatomically and biophysically based modeling framework that can be used to integrate the physiological, anatomical and medical knowledge of the gastrointestinal (GI) system. The second objective is to focus this integrative modeling framework on two major diseases of the GI tract which affect a significant number of people in the United States, namely intestinal ischemia and diabetic gastroparesis. Prior research has shown that recordings of the magnetic field from gastrointestinal electrical activity using multichannel Superconducting QUantum Interference Device (SQUID) magnetometers provides a noninvasive, noncontact assessment of the physiological state of the GI smooth muscle. We will combine multichannel SQUID and cutaneous electrode measurements with anatomically based integrative computer models to investigate the inter- and intra-subject effects of intestinal ischemia and gastroparesis on GI electrical activity noninvasively. We hypothesize that the resulting integration of anatomical and physiological biophysical properties will serve as a basis for a more complete understanding of the gastrointestinal system and will aid in the detection and diagnosis and ultimately, in the treatment of gastrointestinal disorders. This is necessarily a collaborative project that initially involves four groups (the Living State Physics Group at Vanderbilt University, The Department of Surgery at Vanderbilt University, the Department of Physiology and Cell Biology, University of Nevada School of Medicine and the Bioengineering Institute at The University of Auckland) and combines expertise in integrated biophysically based modeling with physiological, clinical and research expertise in the function of the gastrointestinal system.

Grant:	1R01EB003563-01	
Principal Investigator:	RABIN, YOED	DSC
Title:	Developing computerized tools for cryosurgery planning	
Institution:	CARNEGIE-MELLON UNIVERSITY	PITTSBURGH, PA
Project Period:	2004/04/01-2008/03/31	

DESCRIPTION (provided by applicant): Cryosurgery has been known as an invasive surgical technique since 1961, when Cooper and Lee invented the first cryoprobe. In the 1990s, new developments in Joule-Thomson cooling (the cooling effect associated with a sudden relief of a pressurized gas) led to a dramatic decrease in the size of cryoprobes and an increase in the number of cryoprobes that could be used simultaneously. A dozen or more cryoprobes operating simultaneously in a single prostate cryosurgery is already common practice. If localized effectively, one of the primary benefits of using a large number of miniaturized cryoprobes is superior control over the freezing process. Currently, the process of selecting the correct placement of the cryoprobes for a specific procedure is an art held by the cryosurgeon, based on the surgeon's own experience and rules of thumb. Cryoprobes are typically operated in a trial-and-error fashion, until the entire target volume is thought to be frozen. Currently, there are no means to determine the optimal locations for the cryoprobes. Suboptimal cryoprobe localization may leave regions in the target volume unfrozen, may lead to cryoinjury of healthy surrounding tissues, may require an unnecessarily large number of cryoprobes, may increase the duration of the surgical procedure, and may increase the likelihood of post cryosurgery complications, all of which affect the quality and cost of the medical treatment. Computerized planning tools would help to alleviate these difficulties. The "cryoheater," a new device for cryosurgery control has recently been presented by the research team. The cryoheater is a temperature controlled electrical heater. In broad terms, cryoheaters can dramatically increase the ability to control the shape and size of the frozen region, however, to achieve the full benefits of cryoheaters, computerized planning tools for cryoheater localization are necessary. Our goal is to develop computerized planning tools for cryosurgery that are suitable for all available cooling techniques. The proposed research includes: (1) Development of an efficient numerical scheme for bioheat transfer simulations of cyroprocedures, (2) Development of an efficient optimization technique based on a force-field analogy. (3) Development of knowledge-based optimization techniques. (4) Experimental verification of the planning tool. Besides planning, another important application of the proposed tool is the training of cryosurgeons. The proposed tool will provide cryosurgeons with the ability to visualize the 3D volumetric nature of the freezing process. Likewise, it will allow the surgeon to explore the performance of various configurations of cryoprobes and cryoheaters, and observe the defects that would result from each. Such visualization capabilities will provide surgeons with insights into the physics of cryosurgery that are difficult to obtain from physical experiments or surgical practice.

Grant:	1R01DK067198-01	
Principal Investigator:	RODRIGUEZ, LARISSA V	MD
Title:	ADIPOSE DERIVED STEM CELLS FOR TREATMENT OF INCONTINENCE	
Institution:	UNIVERSITY OF CALIFORNIA LOS ANGELES	LOS ANGELES, CA
Project Period:	2004/04/01-2009/03/31	

DESCRIPTION (provided by applicant): Stress urinary incontinence (SUI) is a devastating condition affecting millions of American women. For these patients urinary incontinence is not only an embarrassing condition significantly eroding quality of life, it is also a significant cause of hospitalization. In 1995 the annual cost of incontinence in the United States was estimated to be 26.3 billion dollars. It affects women of reproductive age who are at risk after vaginal deliveries. Its incidence increases with advancing age, making it a major quality of life issue for the elderly. Developing a minimally invasive procedure with high and durable cure rates would have a significant impact on the way physicians treat incontinence and a positive financial impact on health care expenditures. More importantly, it will dramatically improve the quality of life of these patients. With aging there is atrophy of the smooth musculature of the urethra contributing to poor urethral resistance and involuntary loss of urine. Bioengineering new functional tissue in order to increase urethral resistance and improve function has enormous clinical potential for the treatment of stress urinary incontinence. The long-term objective of this application is to apply tissue-engineering techniques exploiting the properties of adult stem cells derived from adipose tissue to develop an effective, minimally invasive treatment for stress incontinence. Our central hypothesis is that human adipose tissue contains a population of pluripotent stem cells capable of differentiating into functional smooth muscle. Specifically, this proposal aims at developing an injectable combination of cells, factors, and matrix to promote the development of vascularized, longlasting functional urethral musculature. The specific aims of this application are: (1) to investigate the ability of human adipose derived stem cells to form functional smooth muscle, (2) to investigate the ability of human adipose derived stem cells to be delivered, survive, and function as normal smooth musculature in the lower urinary tract, (3) to determine the ability of these cells to repair the atrophic nonfunctional urethra of stress incontinence. We will accomplish these aims by evaluating the ability of clonal populations of adipose derived stem cells to differentiate phenotypically and functionally into smooth muscle. Lastly, we will use an animal model of incontinence and decreased urethral resistance to test the hypothesis that these cells can be used to reconstruct a functional urethra as a treatment of stress incontinence.

Grant:	1R01NS048285-01A1	
Principal Investigator:	STANLEY, GARRETT B	PHD
Title:	Nonlinear Thalamocortical Transformations	
Institution:	HARVARD UNIVERSITY	CAMBRIDGE, MA
Project Period:	2004/09/30-2008/06/30	

DESCRIPTION (provided by applicant): The role of the early stages of sensory processing in the brain is to transform signals in the sensory world to provide higher stages of processing with representations that eventually give rise to perception and memory, as well as feedback for motor control. The spatial and temporal transformations by the various stages of processing and the corresponding role of the interplay between sub-populations of excitatory and inhibitory neurons within the pathway in performing these transformations are not well understood. We propose to quantify thalamic and cortical transformations of tactile patterns in the somatosensory pathway through a combination of extracellular recording in the thalamocortical pathway of the rat vibrissa (whisker)system and functional modeling of the corresponding spatial and temporal dynamics, The vibrissa system is an exquisite active sensory modality that rats can use to discriminate between very similarly textured surfaces based on vibrissal exploration alone, an serves as an excellent model system for understanding the transformations of sensory stimuli into thalamic and cortical representations that eventually give rise to perception. The specific aims of this work are to: 1) quantify the transformations induced by the nonlinear temporal dynamics of the thalamus and cortex during the encoding of naturalistic tactile stimuli, 2) quantify nonlinear spatiotemporal transformations in the thalamus and cortex during the encoding of naturalistic tactile stimuli, and 3) determine to what extent these patterns of stimuli can be predicted from known functional properties of the thalamocortical circuit through both biophysically inspired models and estimation techniques focused on nonlinear dynamics. The characterization of these transformations is critical in controlling neuronal function through engineered prosthetic devices in the central nervous system, in order to provide assistance to individuals who have lost sensory function due to disease or trauma, when peripheral devices are not possible or appropriate.

Grant:	2R01AR045748-05A2	
Principal Investigator:	TORZILLI, PETER A	
Title:	Cartilage Cell and Matrix Response to Joint Loading	
Institution:	HOSPITAL FOR SPECIAL SURGERY	NEW YORK, NY
Project Period:	1998/09/30-2009/03/31	

DESCRIPTION (provided by applicant): Little is known about how mechanical loads can initiate the degenerative process in articular cartilage leading to osteoarthritis (OA). Our long-term goal has been to characterize the sequence of physical and cellular events that initiate the degenerative process. To study the degenerative process within a controlled laboratory environment, we used our mechanic explant test system (METS) to model OA by applying an excessive mechanical load (EML) to bovine articular cartilage. Our test system was able to initiate a pathological cascade of events similar to that observed in early stage human and animal OA, including increased enzymes, collagen degradation, proteoglycan loss, and cell death, all localized at the articular surface within the superficial zone (SZ). We have shown that the mechanically-induced damage in the SZ is most-likely cell mediated through the release of three specific metalloproteinases for collagen and aggrecan cleavage, MMPs-1 and 13 (collagenase-1 and 3) and MMP-3 (Stromelysin-1), respectively. However, these results are in contrast to inflammatory-cytokine induced models of OA (IL-1 stimulated) in which aggrecanase-1 and 2 (ADAM-TS4 and 5) were found to initiate the degradation process by aggrecan cleavage. This data leads us to hypothesize that there are two different mechanisms (pathways) of matrix degradation for EML and inflammatory-induced matrix damage. More important, however, is that preliminary results combining both mechanical load and IL-1 stimulation indicate that mechanical loads may inhibit the deleterious effects of IL-1 mediated cell-catabolism by the down-regulation of aggrecanase matrix degradation. We thus further hypothesize that mechanical loads can modulate the degradation process through matrix deformation, that is, by changing the matrix susceptibility to degradation by aggrecanase-1 and 2 and MMPs-1, 3 and 13. The goal of this proposal is to characterize the sequence of degradative events involved in the initiation of matrix damage by EML, to delineate the mechanisms of damage to aggrecan and collagen at the molecular level, to correlate these events and damage to the functional properties (biochemical and biomechanical) of the tissue, and to compare these parameters to those obtained for IL-1 induced damage. If, as we believe, two distinct initiation mechanisms exist, then this would significantly alter how we study each disease process and how we will need to treat them to prevent the progression of the disease.

Grant:	1R01HG003364-01A1	
Principal Investigator:	UGAZ, VICTOR M	PHD
Title:	Novel Convective Flow PCR Thermocycler	
Institution:	TEXAS ENGINEERING EXPERIMENT STATION	COLLEGE STATION, TX
Project Period:	2004/08/18-2007/07/31	

DESCRIPTION (provided by applicant): In order to address the ongoing need for rapid, inexpensive, portable, and reliable genomic analysis equipment, we propose a research effort to demonstrate that convective flow fields can be harnessed to perform the temperature cycling necessary for PCR amplification. A convective flow-based system operates by applying a temperature gradient across a flow network containing the PCR reagent mixture. This temperature gradient establishes a circulatory flow pattern that achieves thermal cycling by continuously transporting reagents through temperature zones associated with denaturation, annealing, and extension reactions. Since the convected reagents are in a continual state of thermal equilibrium with their surroundings, the thermal mass to be heated and cooled consists entirely of the fluid actively involved in the reaction. Consequently, convective flow PCR has the potential to achieve rapid cycling times in reactor volumes ranging from 50 nL to 50 mu L that integrate readily with existing laboratory protocols. By eliminating the need for dynamic external temperature control, a convective flow-based system is capable of achieving performance equal to or exceeding that of conventional thermocyclers in a greatly simplified format, This level of simplicity is a significant departure from previous attempts to construct novel thermocycling equipment, where added complexities often far outweigh any potential performance gains, We propose a research effort targeted at developing a new generation of thermocycling equipment offering improved performance at a significantly lower cost, thereby making PCR practical for use in a wider array of settings. This will be accomplished by achieving the following research Aims, Aim 1: Fully characterize the global 3-D velocity and temperature fields within convective flow reactors using coordinated particle image velocimetry, laser induced fluorescence, and computational simulations. Aim 2: Design, construct, and optimize a series of devices to perform convective flow-based PCR with the ability to achieve high throughput operation and integrate with existing laboratory protocols. Aim 3: Extend the use of convective-flow based systems to satisfy the needs of a variety of thermally activated biochemical reaction systems including RT-PCR, DNA cycle sequencing, and ligase detection.

Grant:	1R01CA102791-01A1	
Principal Investigator:	VINOGRADOV, SERGUEI V	PHD
Title:	Polymer-Nucleotide Complexes with Cytotoxic Activity	
Institution:	UNIVERSITY OF NEBRASKA MEDICAL CENTER	OMAHA, NE
Project Period:	2004/04/01-2008/03/31	

DESCRIPTION (provided by applicant): Effective in chemotherapy of cancer and viral infections nucleoside analogues (NA) are actually 'prodrugs', which must be first converted in vivo into nucleoside 5'-monophosphates and, finally, into the drug's active form, nucleoside 5'-triphosphates. They efficiently terminate DNA synthesis and are cytotoxic for the proliferating cancer cells. However, therapeutic NAs in the form of 5'-triphosphates are considered too unstable as a drug form to be used directly in cancer chemotherapy. Based on preliminary data, the hypothesis being evaluated in this proposal is that encapsulation of 5'-triphosphates of antiproliferative NA in a submicron polymeric carrier with protective and targeting properties will result in a novel therapeutic form of the old drugs. The proposed formulation and delivery system is based on self-assembled polyionic complexes formed between nucleoside 5'-triphosphates and cationic carrier called 'Nanogel'. This carrier consists of a cross-linked network of cationic polyethylenimine and poly (ethylene glycol) or Poloxamer block copolymers. Nanogel loaded with triphosphate nucleotides in aqueous media forms small nanosized particles. Formulated into the particles for systemic administration, active triphosphates of NA can be conventionally stored in freeze-dried form and then readily dispersed before injection. Nanogel can protect triphosphate nucleotides in circulation against enzymatic degradation and drastically increase intracellular transport of anionic nucleotides, which otherwise is not effective. Specific aims of the proposal are to: (1) formulate polymer-nucleotide complexes with increased dispersion stability and enzymatic resistance, (2) Determine whether the polymer-nucleotide complexes can increase the cytotoxic effects of nucleotide analogues, and (3) Examine how the polymer-nucleotide complexes can enhance the systemic therapy of tumors in vivo. A panel of representative NA and cancer cell lines will be studied, and a murine Lewis lung carcinoma model will be used to verify obtained in vitro results. The long circulating polymer-nucleotide complexes can display better tumor accumulation because of the 'enhanced permeability and retention'effect. They can also be modified by vector ligands with affinity to surface receptors on actively proliferating cancer cells in order to enhance selective accumulation of the cytotoxic NA in tumors or metastatic nodes. Application of the drug forms may help to prevent many of the known chemotherapy side effects. Data accumulated in these studies can be directly used for design of better systemic formulations of cytotoxic nucleotide drugs.

Grant:	1R01RR019652-01	
Principal Investigator:	VOLDMAN, JOEL	PHD
Title:	A microscale sorting cytometer for cell-based screens	
Institution:	MASSACHUSETTS INSTITUTE OF TECHNOLOGY	CAMBRIDGE, MA
Project Period:	2004/04/01-2009/03/31	

DESCRIPTION (provided by applicant): We propose the development of a cytometer that combines microscopic imaging with automated sorting. The cytometer addresses two steps in cell-based genetic screens: observation of phenotypes and retrieval of "mutants". Our goal is to be able to sort cells where the sort parameter is based upon the rich information available from microscopy: morphology, intracellular fluorescence, dynamics. Our cytometer is essentially an "active" coverslip that uses an array of reversible addressable traps created with semiconductor technology to hold individual cells in place for observation and release selected cells. The traps use an electrical analogue of optical tweezers--termed dielectrophoresis--to transiently hold the adherent cells in place until they attach to the substrate. After attachment, the cytometer is a passive chip that fits under a conventional upright fluorescence microscope. After live-cell assays have been performed, individual cells in selected traps can be released. The specific aims of our research are to (1) create trapping arrays of increasing complexity, starting with a 4x4 array to verify function, a 20x20 array to characterize sorting, and ending in a 100x100 array for use in an library screen; (2) create the control systems to provide constant temperature, pH, and oxygen tension on-chip coupled with automated microscopy; (3) investigate effects of our cytometer on cell health and perform a phenotype screen.

Grant:	1R01HD044015-01A1	
Principal Investigator:	WALSH, JOSEPH T	PHD
Title:	Spectroscopic Polarized-Light-Based Laparoscopy	
Institution:	NORTHWESTERN UNIVERSITY	EVANSTON, IL
Project Period:	2004/01/01-2007/12/31	

DESCRIPTION (provided by applicant): We propose to improve the visibility of endometrial lesions during laparoscopy. Specifically, we propose the development of a laparoscopic system in which the polarization of the incident and detected light is controlled. The problem is that endometriosis is difficult to diagnose both noninvasively and during laparoscopy. The documented correlation between visual inspection and histological confirmation of suspected lesions is never higher than 65%. We propose to take measurements of light reflected from internal tissue structures thereby obtaining a two-dimensional mapping of the polarization and allowing visualization of the differences that exists between the connective tissue matrix of the lesions and the surrounding normal tissue. The proposed solution takes advantage of several key concepts in tissue optics for the development of a novel laparascope-based system. We will test the safety and efficacy of the system in an animal model, after which the system will be refined as needed. Testing of the system in humans, which is the best model for endometriosis, will be done to demonstrate safety, efficiacy, and provide data for further device improvements.

Grant:	1R01CA106728-01	
Principal Investigator:	WANG, LIHONG	PHD
Title:	Melanoma detection by optical spectroscopy	
Institution:	TEXAS ENGINEERING EXPERIMENT STATION	COLLEGE STATION, TX
Project Period:	2004/09/15-2009/08/31	

DESCRIPTION (provided by applicant): The goal of the proposed research is to develop, test, and validate a preclinical non-invasive optical instrument, based on spectroscopic Oblique Incidence Reflectometry (OIR), for the rapid diagnosis of pre-cancerous and invasive cancerous skin lesions. The underlying hypothesis, which is well supported by our strong preliminary clinical studies, is that quantitative, spatially resolved, optical-spectral (spatio-spectral) images of skin lesions can provide the dermatologist with clinically valuable diagnostic information to supplement his/her clinical judgment about the need for further diagnostic procedures and/or treatments. OIR intentionally breaks the symmetry of diffuse reflectance that exists in its normal-incidence counterpart and, as a result, provides more robust information about the interrogated tissue volume [Wang et al, US Patent, 5,630,423 (1997)]. A preliminary clinical study of 102 skin lesions using a handheld fiber-optic probe showed that the OIR spectroscopic system differentiated cancerous/pre-cancerous skin lesions from benign lesions with better than 95% accuracy. The applicants have identified the key diagnostic physiological parameters to be (1) the oxygen saturation of hemoglobin, (2) the concentration of total hemoglobin, and (3) the cell-nuclear size. The engineering goal of this proposal is to improve the prototype OIR system to measure skin lesions in real-time and process the data optimally for accurate lesion classification. The mechanistic goal is to identify and quantify the tissue and cellular factors that are responsible for the diagnostic optical features. The clinical goal is to provide accurate, objective, and real-time diagnoses for benign, pre-cancerous, and malignant lesions that are difficult to diagnose clinically. The ultimate societal impact is to improve patient care, save lives, and reduce health care costs. The Specific Aims are listed as follows. 1. To improve instrumentation. 2. To establish a statistically significant database. 3. To develop and validate a diagnostic algorithm. 4. To identify the pathophysiologic parameters responsible for the diagnostic optical features.

Grant:	1R01GM071321-01A2	
Principal Investigator:	WATERS, CHRISTOPHER M	PHD BIOMEDICAL ENGINEERING & PHYS
Title:	ABC Transporters in CNS Penetration of Camptothecins	
Institution:	UNIVERSITY OF TENNESSEE HEALTH SCI CTR	MEMPHIS, TN
Project Period:	2004/07/01-2008/06/30	

DESCRIPTION (provided by applicant): Tightly regulated cellular barriers limit delivery of pharmacological agents to children with primary tumors in the central nervous system (CNS). The physiological regulation of the blood-brain barrier (BBB) and the blood-cerebrospinal fluid (CSF) barrier (BCB) and the specificity for excluding or allowing drug transport are not well understood. Camptothecin analogs such as topotecan and irinotecan are used to treat children with primary CNS tumors, but the CNS distribution of these drugs is widely different. The discovery of Pglycoprotein (P-gp) and other ATP-binding cassette (ABC) transport proteins has challenged the view that only passive diffusion and physicochemical properties control drug distribution. The major hypothesis of this proposal is that the distribution of camptothecin analogs between the blood, brain tissue, and the CSF is largely controlled by drug efflux transporters, including P-pg, multi-drug resistance protein 4 (MRP4), and the breast cancer resistance protein (BCRP or ABCG2). We seek to understand how drug distribution is controlled at the BBB and the BCB. In Aim 1 we will develop tissue engineered models of the BBB and BCB to investigate drug transport in vitro. Cells transfected with different ABC transporters will be used to determine the specificity of transporters for camptothecin analogs. Rat brain endothelial cells and astrocytes will be co-cultured on filter inserts and in hollow fiber cartridges to examine vectorial drug transport in the absence and presence of shear stress. Mathematical models will be used to evaluate transport parameters from experimental data. In Aim 2 we will determine the mechanisms of camptothecin analog transport in vivo using immunohistochemistry to determine the anatomical distribution of ABC transporters, and microdialysis measurements of brain and CSF drug concentrations in knockout mice deficient in MRP4 and ABCG2. In Aim 3 we will determine if ancillary drug therapy alters expression of drug metabolizing enzymes or ABC transporters in the CNS. In Aim 4 we will determine the expression and location of ABC transporters in primary CNS tumors and relate that to intracranial penetration of campothecin analogs and antitumor response. Our long-term goals are to understand the regulation of drug transport in the CNS, to develop in vitro and in vivo models to test camptothecin analogs, and to define and quantify parameters that can be used to compare drugs and ultimately improve therapy for children with primary CNS tumors.

Grant:	2R01HL064981-05A1	
Principal Investigator:	WATERS, CHRISTOPHER M	PHD BIOMEDICAL ENGINEERING & PHYS
Title:	Biomechanics and Wound Healing in Lung Epithelial Cells	
Institution:	UNIVERSITY OF TENNESSEE HEALTH SCI CTR	MEMPHIS, TN
Project Period:	1999/09/07-2009/02/28	

DESCRIPTION (provided by applicant): A recent randomized clinical trial by the Acute Respiratory Distress Syndrome (ARDS) Network demonstrated a 22% reduction in mortality in patients by simply reducing the tidal volume for mechanical ventilation from the conventional setting of 12 ml/kg to a lower setting of 6 ml/kg. This dramatic decrease in morbidity and mortality has stimulated significant interest in the mechanisms of ventilator-induced lung injury (VILI) and the development of lung protective strategies. Rapid repair of the alveolar epithelium following injury is crucial for restoration of barrier function and gas exchange, while restoration of the airway epithelium is important to prevent fibroproliferation and occlusion of the airways and to reduce the possibility of infection. The present proposal is designed to address the hypothesis that increased mechanical tension inhibits healing of injured pulmonary epithelium. This is important because epithelial repair may be inhibited in patients with acute lung injury in whom overdistention of lung epithelium occurs during positive pressure mechanical ventilation. The majority of recent studies examining ventilator-induced lung injury have focused on how mechanical forces activate proinflammatory pathways or induce additional injury. Our study explores the role of mechanical forces during the reparative phase following injury. Mechanisms by which mechanical strain inhibits wound repair will be examined using an in vitro model of alveolar and airway epithelial cells cultured on elastic substrates. The hypothesis that mechanical strain alters localized levels of mechanical tension that drive wound repair will be tested. Atomic force microscopy will be used to measure localized cell stiffness. Changes in cytoskeletal remodeling, microtubule growth, focal adhesions, and other structural components of wound healing in response to mechanical strain will be examined. Mechanotransduction pathways involving Rho GTPases and cytoskeletal remodeling during wound repair will be investigated. Since there is some uncertainty regarding the degree of deformation of airways in vivo, mechanical strain will be measured in mechanically ventilated rats using microfocal X-ray imaging of tantalum-coated airways. The proposed work will provide a foundation for understanding the role of mechanical forces and ventilator settings in epithelial repair mechanisms following lung injury.

Grant:	1R01HL077683-01	
Principal Investigator:	WEISS, JEFFREY A	PHD
Title:	Angiogenesis and the Extracellular Matrix	
Institution:	UNIVERSITY OF UTAH	SALT LAKE CITY, UT
Project Period:	2004/01/01-2008/12/31	

DESCRIPTION (provided by applicant): The broad objective of this Bioengineering Research Grant is to study biomechanical interactions of angiogenic microvessels with the extracellular matrix (ECM) on the microscale level. We will answer the following questions: How does angiogenesis influence global and local ECM material properties and ultrastructure? Is local angiogenic sprouting correlated with the stress state as predicted by computational mechanical modeling, MMP expression and ECM ultrastructure? Does mechanical conditioning of vascularized constructs influence angiogenic sprouting? To answer these questions, we will develop and apply novel experimental and computational techniques to study a 3D in vitro angiogenesis model. In the first specific aim, we will develop techniques to simulate the microscale biomechanical behavior of vascularized collagen gels using the Material Point Method (MPM), using volumetric confocal images as the basis for generating the geometry of the computational domain. In Aim 2, methods will be developed to nondestructively measure collagen gel ultrastructure, microvessel geometry and emission spectra using spectrofluorimetry and multiphoton fluorescence microscopy. In Aims 3 and 4, these highly novel approaches will be combined with traditional approaches for experimental measurements of biomechanical behavior, gene expression and protein expression to examine the mechanisms that are responsible for alterations in ECM material properties during angiogenesis. Finally, we will examine the effects of mechanical conditioning on microvessel sprouting and growth. The proposed experiments will provide an information base on the magnitudes and frequencies of forces that most influence the angiogenic vessel. A better understanding of the relationship between angiogenic vessels, the surrounding ECM structure, and the mechanics of the tissue undergoing angiogenesis will provide the basis for improved control of tissue vascularization in both native tissues (e.g., repairing ischemic tissue) and tissue engineered constructs.

Grant:	2R01AR047369-05	
Principal Investigator:	WEISS, JEFFREY A.	PHD
Title:	Origins of Elasticity/Viscoelasticity in Knee Ligaments	
Institution:	UNIVERSITY OF UTAH	SALT LAKE CITY, UT
Project Period:	2000/09/01-2009/08/31	

DESCRIPTION (provided by applicant): The knee ligaments are short bands of tough fibrous connective tissue that guide normal joint motion and restrict abnormal joint movement. Although the injury and healing of knee ligaments have been topics of extensive study, fundamental information about the relationship between the ultrastructure of the tissue and its continuum level mechanical behavior is severely lacking. The decorin-based proteoglycans play an important role in collagen fibrillogenesis and may be important determinants of the material behavior of connective tissues. The objectives of this research are to investigate the roles of decorin-based proteoglycan crosslinks and fluid flow in the elastic and viscoelastic material behavior ligaments. The hypotheses to be addressed are 1) decorin-based crosslinks control the resistance of ligament to tensile loading transverse to the fiber direction and shear loading along the fiber direction by stretching during relative movement of the collagen fibrils; 2) a transversely isotropic hyperelastic constitutive model that incorporates structural information regarding decorin crosslinks and collagen fiber crimp will describe and predict the elastic material behavior of human knee ligaments; 3) The viscoelastic material behavior of ligament is due to fluid movement. These hypotheses will be addressed through a series of aims that combine experimental measurements from the molecular level to the continuum level. The results of this study will have important implications for understanding the fundamental role of the small proteoglycans and fluid flow in the viscoelastic behavior of fibrous connective tissues, and will help to understand the phenotypes associated with disease states that are related to deficiencies in the small proteoglycans.

Grant:	1R01DC006458-01A1	
Principal Investigator:	WIET, GREGORY J	MD
Title:	Virtual Temporal Bone Dissection	
Institution:	CHILDREN'S RESEARCH INSTITUTE	COLUMBUS, OH
Project Period:	2004/07/15-2009/06/30	

DESCRIPTION (provided by applicant): Otologic disease accounts for an estimated 8 billion in health care costs annually in the United States. Training health professional charged with the surgical treatment of such disease requires 5 to 7 years of training at an annual cost of $76,000 each. Currently, this requires mock surgical procedures using cadaver material (temporal bones) and apprentice type training on real patients in the operating room. The expected application of this research is to provide an adjuvant (auxiliary) environment to learning the surgical treatment of otologic disease. Our long-term hypothesis is that simulation technologies can increase efficiency in training and raise proficiency of the practitioner in a safe and cost effective manner. For this specific project, our focused hypothesis is that a virtual environment for temporal bone dissection is equivalent to training with cadaveric temporal bone dissection in the anatomy laboratory. The broad, long-term objectives of this work are to further develop and validate a robust, realistic virtual environment for temporal bone dissection. Specifically, this work will evaluate the efficacy of emerging simulation technologies compared to traditional methods of temporal bone dissection for training otologic surgeons. This work will extend the functional use of the system (as developed under a previous R21) through increased realism, the integration of new high and ultra high-resolution multimodal image data sets, and through a multi-center national trial. In its current state, the system has met with overwhelming support from a number of academic institutions, which have currently committed to further collaborative development and validation studies.